The Succession of Forensic Beetles on Exposed and Wrapped Carcasses During Winter and Summer in Khartoum State

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A Thesis Submitted in partial Fulfilment of the Requirements for the Degree of M.Sc.in Medical Entomology and Vector Control

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October 2014
DEDICATION

Father
Mother
My sisters
My Friends I would like to dedicate this work to
My family
Friends
And
Colleagues

Sarah
ACKNOWLEDGEMENTS

I am greatly indebted to my supervisor Professor El Amin El Rayah Mohamed, Department of Zoology, Faculty of Science, University of Khartoum for his support, continuous encouragement, patience, guidance and for reading the draft manuscript. And I am very grateful to all my Colleagues for their help and cooperation in this work. Grateful thanks are due to the Department of Zoology for their generous hospitality.
ABSTRACT

Forensic entomology is the study of arthropods associated with dead bodies. Arthropod succession studies have been successfully used to estimate the postmortem intervals. Most research in forensic entomology has focused on flies more than beetles (Coleoptera).

Coleoptera is the second largest order of forensic interest, with several necrophagous representatives, most being predators but their feeding habit may change between larval stages and adulthood.

This research is aimed to determine the decomposition stages and Coleoptera succession on different carcasses (rabbits, pigeons and fishes) and test the influence of wrapping on the decomposition of the carcasses and the Coleoptera succession during winter and summer seasons.

Freshly killed rabbits (*Oryctolagus cuniculus*), fishes (*Oreochromis niloticus*) and pigeons (*Columba livia domestica*) were used either exposed or wrapped with cloth (three layered sheets of dabalan) and tightly tied by a piece of cloth at both ends.

Six sets of these animal carcasses were used in each trial (two exposed rabbits, two wrapped rabbits, two exposed fishes, two wrapped fishes, two exposed pigeons and two wrapped pigeons). Daily observations were made and beetles presence was regularly recorded.

The coleopterans observed in this study, were represented by four families namely (1)Staphylinidae (rove beetles), (2) Histeridae (clown beetles),(3) Cleridae (red-legged ham beetles), and (4) Dermestidae (skinbeetles), seven genera and species of these families were recorded:

The results and observations also showed that: (1) Carcasses in summer decayed much faster than those in winter. (2) Exposed carcasses decayed faster than wrapped ones. (3) Beetles recorded higher frequencies in exposed carcasses than wrapped carcasses, and those in summer were more than those in winter. That means the decomposition process and the Coleoptera succession were affected by seasons (temperature) and wrapping.
المستخلص

علم الحشرات الجنائي أو الشرعي يهتم برسمة المفصلات ذات الصلة بالجثث. الأبحاث التي
إهتمت بدراسة تتابع أوتعاب المفصلات نجحت في تحديد الفترة بين حدوث الوفاة و اكتشاف
الجثة. معظم الدراسات في هذا المجال ركزت على دراسة الذباب أكثر من الخناص.

رتبة غديميات الأجنحة أو الخناص هي ثاني أكبر رتبة من الحشرات ذات الأهمية الجنائية أو
الشرعية. العديد منها أكلات جيف ومعظمها مفترسات ولكن طريقة غذائها قد تختلف في مختلف
أطراف حياتها من طور اليرقة إلى الطور البالغ.

هذه الدراسة حاولت تحديد مراحل التحلل وتعاقب الخناص على جثث حيوانات مختلفة
(أرانب، أسماك، وحمام) وأيضاً اختبار تأثير الغطاء على عملية التحلل وتعاقب الخناص في كل
من موسم الشتاء والصيف.

تم استخدام جثث حيوانات حديثة الموت. أرانب من نوع (أوريكتولاكون كوبنكولس).
أسماك من نوع (أوريوكروست ذيلوتكس) وحمام من نوع (كولومبا ليفيا دومستكا). تم وضع هذه
الجثث إما مكشوفة أو مغطاة بثلاث طبقات من قماش الدبلاين مربوط ربطاً محكماً عند طرف
الجثة.

ست مجموعات من الجثث استخدمت في كل تجربة (أرناب مكشوف، أرناب مغطية،
سمكタン مكشوفات، سمكタン مغطية). حماتان مكشوفتان وحمامتان مغطيتان). وقد تم أخذ
الملاحظات وتسجيل حضور الخناص يوميا.

تغديميات الأجنحة التي تم رصدها تتمثل في أربع عوامل وهي: (1) الخناص الطواف أو
الجوالة. (2) الخناص الصليبة. (3) الخناص حمراء الأرجل. (4) خناص الجلود. تم رصد سبعة
أجناس وأنواع من هذه العوائل. الخناص حمراء الأرجل (نوروبيا ريفيس). خناص الجلود
(ديمستن ماكولاسس و ريسا فسيولي). الخناص الصليبة (هستر. إيوبلوتس و زيرويرن)
والخناص الجوالة (فولنس).

النتائج والملاحظات التي تم الحصول عليها أوضحت أيضاً أن:
(1) الجثث في فصل الصيف تتحلل بسرعة أكبر منها في فصل الشتاء.
(2) الجثث المكشوفة تتحلل بسرعة أكبر من المغطاة.
(3) الخناصس تتردد على الجثث المكشوفة بأعداد أكبر من المغطاة. كما إنها توجد في فصل الصيف بأعداد أكبر منها في فصل الشتاء.

هذا يعني أن عملية تحلل الجثث وتعاقب الخناصس (غمديات الأجنحة) تتتأثر بتغير الفصول (درجة الحرارة) كما تتتأثر بالغطاء.
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INTRODUCTION AND LITERATURE

REVIEW

1. Definition

Forensic entomology is the branch of forensic science, in which information about insects is used to draw conclusions when investigating legal cases related to both humans and wildlife. This term may be expanded to include other arthropods. Insects can be used in the investigation of a crime scene both on land and in water (Anderson, 1995; Erzinçlioğlu, 2000; Keiper and Casamatta 2001; Hobischak and Anderson 2002; Oliveira-Costa and de Mello-Patiu, 2004). The majority of cases where entomological evidence has been used are concerned with illegal activities, which take place on land and are discovered within a short time of being committed.

Within the framework of a judicial inquiry following the discovery of a cadaver, the determination of time of death is a very important issue for the legal authorities (police officers, magistrates, coroners, etc.). Such estimation is more difficult to establish when the cadaver has reached an advanced stage of decomposition. In this case, the entomological evidence can be one of the few sources to make such estimation. However, the process of decay of organic material is highly complex and numerous interrelated factors influence it: macroclimate, microclimate, availability and accessibility of insects to the carcasses (Braack, 1981).

Medicolegal forensic entomology includes arthropod association with felonies such as murder, suicide and rape. In its most practical form,
information dealing with carrion insect ecology has been used to estimate the minimum elapsed time since death in homicide cases.

Insects are the primary fauna associated with carrion (Goff and Odom 1987; Payne 1965; Tantawi et al. 1996). It is known that there is an assemblage of insect species that are attracted to decomposing animal remains and play an active role in the decay process (Anderson and Cervenka 2002; Lord 1990). Certain species in the orders Diptera and Coleoptera represent the majority of the total necrophagous fauna found on carrion (Greenberg 1991; Payne 1965).

Many of these insects have been classified according to their ecological role in carcass decomposition. Typically, carrion is rapidly invaded first by necrophagous flies in the families: Calliphoridae, Sarcophagidae and Muscidae. This results in the presence of huge numbers of fly eggs and larvae, thereby providing an abundant food supply for predacious beetles such as Silphids, Staphylinids and Histerids. As the carcass decomposes and food resources become depleted, it becomes attractive to different insect species for feeding and reproduction. Species in the families: Piophilidae, Cleridae and Nitidulidae are typically associated with the carcass during the later stages of decay and the Dermestidae are attracted to the dry remains (Payne and King 1970; Smith 1986; Tantawi et al. 1996). Since many of these carrion insects are attracted during these different stages of decomposition, it has been shown that carrion is invaded by certain insect species in a typical sequence or succession (Anderson and VanLaerhoven 1996; Bourel et al. 1999; Early and Goff 1986; Goff and Flynn 1991; Haskell et al. 1989; Lord and Burger 1984; Payne 1965; Tantawi et al. 1996; Tullis and Goff 1987).
Many factors can affect insect succession and the decomposition of carrion. Factors such as temperature, season, time of day, accessibility and physical position of a carcass, size and type of carcass, vertebrate scavengers, insect abundance and the biology and geographical distribution of the necrophagous insects, can influence the time of arrival and the duration of stay of insects on the carrion (Anderson 2000; Dillon 1997; Hall and Doisy 1993; Payne 1965; Tullis and Goff 1987). These many factors make it necessary to study insect succession on carrion in different regions and under different conditions.

The succession of insect species on carrion varies according to temperature, habitat and geographic location (Dillon (1997), Goff (1991), Goff and Flynn (1991)).

2. Rate of Decomposition

There are three recognizable processes in corpse decomposition. These are autolysis, putrefaction and skeletal bone decomposition (diagenesis). In autolysis, a process of natural breakdown, the cells of the body are digested by enzymes, including lipases, proteases and carbohydrases. This process can be most rapid in organs such as the brain and liver (Vass, 2001). A ‘soup’ of nutrients is released which forms a food source for bacteria. Putrefaction is the breakdown of tissues by bacteria.

As a result, gases such as hydrogen sulphide, sulphur dioxide, carbon dioxide, methane, ammonia, hydrogen and carbon dioxide are released. Alongside this, anaerobic fermentation takes place when the volatiles propionic and butyric acids are formed. The body undergoes active decay, in which protein sources are broken down into fatty acids by bacteria (Vass, 2001). Fatty acids and such compounds as skatole, putrescine and cadaverine are significant members of these decomposition products.
(although Vass et al., 2004, commented on their absence from recovered volatiles from buried bodies).

The body can be allocated to one of five recognizable post mortem conditions, which can be linked to the eight waves of arthropod colonization proposed by Mégnin (1894). No distinction from one stage to the next is obvious and Gaudry (2002), on the basis of 400 cases, considers Mégnin’s first two waves to be one. Although no stage has a fixed duration, each stage can be associated with a particular assemblage of insects. The profiles of insects would appear to be universal, although the majority of research on this aspect has, until recently, been undertaken in North America (Hough, 1897; Easton and Smith, 1970; Rodriguez and Bass, 1983; Catts and Haskell, 1990; Mann, Bass and Meadows, 1990; Goff, 1993; Dillon and Anderson, 1996; VanLaerhoven and Anderson, 1999; Byrd and Castner, 2001). These stages of post mortem changes are:

- **Stage 1: Fresh stage.** This stage starts from the moment of death to the first signs of bloating of the body. The first organisms to arrive are the blowflies (the Calliphoridae).

- **Stage 2: Bloated Stage.** Breakdown of the body continues because of bacterial activity, or putrefaction, and this is perhaps the easiest stage to distinguish.

  Gases causing the corpse to bloat are generated through metabolism of nutrients by anaerobic bacteria. Initially the abdomen swells but later the whole body becomes stretched like an air-balloon. At this stage more and more blowflies are attracted to the body, possibly in response to the smell of the breakdown gases. Vass et al. (1992, 2004) studied the odours emanating from dead bodies that were both resting on the surface and had been buried. Their work provides clarification of the identity of some of
these gases and the information supplements that provided by Mégnin (1894); Hough (1897) and Smith (1986).

Rove beetles (Staphylinidae) may be attracted to the body at the bloat stage because of the ‘ready meals’ of eggs and maggots. These and other predators can affect the interpretation of the range of insects and insect life stages present as they feed on larvae or remove puparia (Smith, 1986).

• **Stage 3: Active decay stage.** This stage is recognizable by the skin of the corpse breaking up and starting to slough from the body. This sloughing allows the decomposition gases to escape and so the inflation of the body gradually subsides as putrefaction continues. In the later stages of putrefaction fermentation occurs and butyric and caseic acids are generated. This is followed by a period of advanced putrefaction, which includes ammoniacal fermentation of the body, to which a different cohort of insects are attracted. These include the silphid beetle *Nicrophorus humator* (Gleditsch) the histerids *Hister cadaverinus* (Hoffmann) and *Saprinus rotundatus* Kugelann, and the muscid fly *Hydrotaea capensis* Wiedeman (= *Ophyra cadaverina* Curtis).

• **Stage 4: Post-decay stage.** In the later stages of decay, all that remains of the body are skin, cartilage and bones with some remnants of flesh including the intestines. Any remaining body tissue can be dried. The biggest indicator of this stage is an increase in the presence of beetles and a reduction in the dominance of the flies (Diptera) on the body.

• **Stage 5: Skeletonization.** At this stage the body is only hair and bones. No obvious groups of insects are associated with this stage, although beetles of the family Nitidulidae can, in some instances, be
found. The body has clearly reached its final stage of decomposition. Any further breakdown is best described in terms of the decay of individual components of the body, such as the bones of the feet and legs, the skull and the ribs.

The zoogeographical region, country and type of terrain within a given country will affect the composition of the faunal succession and the rate of decomposition. (Smith 1986)

3. Wrapped Remains

Remains, whether whole or dismembered, are frequently found wrapped in some material. This may be an effort to disguise the remains, to facilitate handling, or to prevent bleeding onto a carpet or vehicle. The type and extent of the wrapping may affect the insect colonization pattern of the remains, but possibly provided protection from predators and the elements. More secure wrapping may delay insect colonization.

The wrapping did influence decomposition rate, and in particular, the rate of drying. Wrapped carcasses did not enter typical dry remains stage, but rather, stayed moist, although there was no change in insect succession of the carcasses. The distribution of the maggot masses was impacted by the sheet, as wrapping appeared to facilitate maggot mobility (Kelly 2006; Kelly et al. 2008, 2009), (Jason and James 2001).

The wrapped carcasses remained moist. This suggested that the wrapping allowed little evaporation and/or slowed the draining of the decomposition fluids onto the ground, and thereby carcasses were kept moist. This caused the wrapped carcasses to remain in the advanced decay stage for significantly longer time. This observation would need to be taken into consideration when dealing with human remains in cases
where those remains are in the advanced stage of decay. The insect succession did not change although the physical appearances of the carcasses were different. (Kelly 2006).

4. Beetles as indicators of time of death

Insects are used in forensic investigations primarily to develop an estimate of minimum post mortem interval (PMI$_{\text{min}}$). These estimates can be based on the duration of the immature stages of the insects found on a corpse or on the community composition of insects on the corpse (Byrd and Castner 2001). The duration of the immature stages is generally longer in Coleoptera than in Diptera, which means that Coleoptera are useful to estimate PMI$_{\text{min}}$ not only during early decomposition, but also in later stages of decomposition. In addition, many beetles utilize corpses in advanced decomposition and can be used to estimate PMI$_{\text{min}}$ by analyzing the community present on a corpse (Smith 1986). In these cases many fly larvae have already left the corpse, leaving mostly Coleoptera from which to make estimates.

The most precise method of estimating PMI$_{\text{min}}$ using insects is to use models based on development of immature stages (Higley and Haskell 2001). These models can either use size as a surrogate for age or use physiological age by identifying developmental landmarks. The latter models are less biased and more precise (Dadour et al. 2001), as they measure actual age, and not size, which can be affected by many factors other than age (Villet et al. 2009). Development of flies has been investigated extensively and refined models for various species are available (Grassberger and Reiter 2001, 2002; Higley and Haskell 2001; Villet et al. 2006; Richards et al. 2008). Statistically robust models for
coleopteran development are not as common (Midgley and Villet 2009a) and so data for most species should be interpreted with caution. This is not to say that all data should be disregarded, but further study is required to develop statistically robust models. The development of *Thanatophilus micans* has been thoroughly modelled (Midgley and Villet 2009a) and shows that with more research, development of Coleoptera can be a useful tool for forensic entomologists. The models produced for the developmental landmarks of *T. micans* not only meet the minimum statistical requirements for regression modelling (Richards and Villet 2008), but have coefficients of determination greater than 0.98 for all post-hatching stages (Midgley and Villet 2009a). This shows that beetle development is predictable and, coupled with the rapid location of corpses, shows that at least *T. micans* and probably other sexton beetles (Silphidae) are reliable forensic indicators.

In many cases live insects are not available for PMI$_{\text{min}}$ estimation, usually because they are collected by non-specialists (Lord and Burger 1983). In such cases the length, width or mass of the collected specimens is the only reliable measure for estimating PMI$_{\text{min}}$. Size-at-age data is not available for most forensically relevant beetle species, with the exception of *T. micans* (Midgley and Villet 2009a). Change in specimens’ sizes during storage is a well known fact in forensic entomology (Lord and Burger 1983; Adams and Hall 2003; Amendt *et al.* 2007; Midgley and Villet 2009b) and this must be considered when using developmental models based on length. The killing method used to preserve samples has an effect on the change during storage and must also be considered. For fly larvae, killing with ethanol is not recommended, as significant changes in length occur (Tantawi and Greenberg 1993; Adams and Hall 2003). This is not the case with beetle larvae: killing with ethanol causes the least change in length of silphid larvae (Midgley and Villet 2009b)
and is therefore the most suitable preservation method. This is because beetle larvae have extensively sclerotized exoskeletons, and so are more rigid than fly larvae. Similarities and differences between Coleoptera and Diptera must be considered when samples are taken at a crime scene to obtain accurate estimates of PMI_{min}.

An advantage of estimating PMI_{max} (maximum post mortem interval) from beetle larvae is that they are solitary and furtive, while maggots aggregate into maggot balls or maggot masses. This results in beetle larvae experiencing temperatures close to ambient, which simplifies the application of thermal accumulation models of development. Blowfly larvae in maggot masses collectively generate enough heat to warm themselves as much as 25°C above ambient temperatures. Accounting for this while estimating a PMI_{max} is a source of error that can be avoided by using both flies and beetles in a given estimate.

The use of development to estimate PMI_{min} is not limited to the primary consumers of decaying corpses. Parasitoids and predators, such as rove beetles (Staphylinidae) and clown beetles (Histeridae), of these species can also be used, as their larvae are also obligate corpse dwellers. The precision and accuracy of these estimates may be decreased because they are subject to the developmental variability of the necrophilous parasite or predator in addition to that usual in the necrophagous species.

The latter may even be modified by parasitoids. Fly pupae of several species and families can be parasitized by species of Aleochara (Staphylinidae) (Gauvin 1998; Ferreira de Almeida and Pires do Prado 1999). Aleochara is however a large and diverse genus, with between 300 and 400 species (Maus et al. 2001), many of which are geographically localized or do not parasitize necrophagous Diptera. Identification of locally relevant species and the generation of developmental models for
these species are critical before *Aleochara* can reach its potential in estimating the $\text{PMI}_\text{min}$.

### 4. Coleoptera

Coleoptera is the second largest order of forensic interest, with several necrophagous representatives, most being predators but their feeding habit may change between larval stages and adulthood. The species of Coleoptera increase in number both of individuals and species during advanced stages of decomposition in open environment and are absent or less represented indoors (Goff 1991). Beetles are encountered in great numbers during the faunal succession process; moreover their biological traits may be used to estimate the *post mortem* interval.

According to Smith (1986) the families of Coleoptera of forensic interest are: Carabidae, Hydrophilidae, Silphidae, Leiodidae, Staphylinidae, Histeridae, Cleridae, Anthicidae, Dermestidae, Nitidulidae, Rhizophagidae, Ptinidae, Tenebrionidae, Scarabaeidae, Geotrupidae and Trogidae.

Many beetles are specifically associated with carrion, the majority is probably predators and only a few are true carrion feeders. The feeding habits of adults and larvae may also differ in a particular species and are discussed more fully under each family below.

### Taxonomy

One of the key needs of a forensic entomologist is a method of identifying insects found on corpses. Necrophagous Coleoptera found on older or dryer corpses can be identified using the keys provided in the stored product literature, such as Hinton (1945) and Gorham (1987).
Works more specifically about stored product pests are also useful. Dermestid adults (Mroczkowski 1968; Peacock 1993) and larvae (Rees 1947; Adams 1980; Zhantiev and Volkova 1998, 1999) are common on corpses and most are cosmopolitan, making identification easier. Cleridae are represented by a few species of *Necrobia*, particularly *N. rufipes* that are widespread stored product pests, which simplify their identification (Smith 1986; Gorham 1987; Rajendran and Hajira Parveen 2005). Ptinidae are also found on dessicated bodies, and can be identified using Brown (1940), Harney (1993) or Irish (1999) guides.

For exclusively Necrophagous species, such as Silphidae, Staphylinidae and Histeridae, identification is not as easy because many of them are neither cosmopolitan nor pestilent. Their identification therefore depends on the taxonomic advancement of the broad geographic area in which the corpse is located. African and Australian Silphidae can be identified using Schawaller (1981, 1987) and Peck (2001), and the Afrotropical Trogidae using Scholtz (1980, 1982) and van der Merwe and Scholtz (2005); these works allow easy identification of these species. With a little more effort, one can identify many genera of Histeridae using Caterino and Vogler (2002) and all tribes of Staphylinidae using Solodovnikov and Newton (2005); Catalogues of local necrophilous beetles, such as the Turkish species found by Özdemir and Sert (2008), can provide easy identification of beetles in a given area, but should be used with caution outside of the geographic range treated.

**Biology**

Once the species on a corpse are identified, information will be needed about their biologies. Despite the fact that little research on beetles has been focused explicitly on forensics (Williams and Villet 2006), notes and data on the development of several coleopteran species
can be found in various publications. The more common and widespread species are often stored product pests, and significant research has been carried out in efforts to control these species. A good example of this is *Dermestes maculatus*, a cosmopolitan pest of stored products (Rajendran and Hajira Parveen 2005) that is common on mummified remains (Schröder et al. 2002). It is important to analyse the beetle community as a whole, as not all beetle species will oviposit immediately after death. This delay means that the precision of an analysis using only one species will be reduced. When the biology of *Dermestes* spp. is used to estimate PMI$_{\text{min}}$, it becomes clear that adjustments may need to be made to the estimate because key factors modifying growth in *Dermestes* spp. are moisture content in food (Scoggin and Tauber 1951) and relative humidity (Coombs 1979).

It is therefore important to adjust PMI$_{\text{min}}$ estimates based on the development of these species to account for relative humidity and dietary moisture. In cases where dietary moisture remains extremely high or low during decomposition, the development of *Dermestes* spp. will not give unbiased PMI$_{\text{min}}$ estimates as development will not occur at normal rates (Schröder et al. 2002). In these cases other species found on the corpse should be used in conjunction with *Dermestes* spp. for PMI$_{\text{min}}$ estimates, such as *T. micans* and other Silphidae during early decomposition and *N. rufipes* and *Aleochara* spp. in advanced decomposition. By assessing the development of as many species as possible, a crossvalidated view of the community can be obtained for analysis, making oviposition and biological variations less important and giving a more unbiased and precise PMI$_{\text{min}}$ estimate.
The life stages of the beetles

Beetles are insects which also show complete metamorphosis. The term for this is *holometabolous*. To become adults, they too pass through an egg stage, three to five larval stages depending on species, and a pupal stage. Coleopteran eggs tend to be oval, spherical or spheroid in shape and are usually considered very similar, irrespective of family. Beetles usually bury themselves in the ground, or in a specially constructed chamber, when they pupate. Less detailed information is available about beetle life cycles than is known about the Diptera.

The length of the life cycle will vary, depending upon the family and species of beetle. Development through the complete life cycle, from egg to adult (imago) can take 7–10 days in rove beetles (Staphylindae), whereas in the ground beetles (Carabidae) completion of the life cycle to the adult stage may take a year and the adults may live for 2–3 years. In some species the number of instars in the larval stage is not fixed, but is dependent on environmental conditions. In Dermestidae, for example, there may be as many as nine instars (Hinton, 1945). Usually, however, there is only one generation of beetles per year. Smith (1986) indicates that the length of the pupal stage of *Dermestes* sp. can last between 2 weeks and 2 months and that these beetles can overwinter (enter *diapause*) in a pupal chamber if the weather is not suitable or it is late in the season.

The problem of lack of a ready morphological distinction between larval instars in beetles means that other methods to distinguish the instar are needed.

Ecology of skin, hide and larder beetles (Dermestidae)

Several species of dermestid beetles have been shown to colonize a dead body. These include *Dermestes ater* DeGeer, *Dermestes maculatus*,

...
Dermestes lardarius and Dermestes frischii (Kugelann) (Centeno et al., 2002). Dermestes maculatus will be used as an example of the response of dermestids to a corpse, since this species is well researched because of its role as a stored product pest.

Dermestes maculatus growth from egg to adult can take 20–45 days, although the speed of development depends on the temperature of the habitat. The larvae have characteristic hairs on their body segments and are referred to colloquially as ‘woolly bears’. These hairs occur in tufts at the end of the body or along the sides of each segment and, according to Hinton (1945), can be moved or vibrated when the larva is being threatened.

Adult dermestids show a negative response to light (negative phototaxis) and will, when touched, readily ‘play dead’ (show thanatosis). Dermestids will happily exist in darkness as larvae. However, when food is in short supply, the beetles have been known to walk or fly away from the current food source towards a light source. These habits mean that they can be kept in the dark but need a reliable food source and pupation site to successfully complete their life cycles.

On a body on which no live specimens of dermestids remain, their frass provides evidence of forensic significance; being an indicator that this species was formerly present. Frass has a characteristic twisted shape and is white in colour. It comprises undigested food, which is encased in peritrophic membrane. Where frass alone is present, it may reflect dermestid activity for a period of time between 1 month and 10 years. Indeed, Catts and Haskell (1990) recorded frass originating from dermestids on 10 year-old mummified bodies retained in a house by a solicitous, but criminally culpable, relative.

Ecological conditions appear to determine whether dermestid species will be present. Arnoldos et al. (2005) showed that the coleopteran profile
in south-eastern Spain varied in both distribution and abundance throughout the year. They recorded few dermestid species in the earliest stages of corpse decomposition in spring and summer. Subsequently, numbers of the dermestid species increased as the remains began to dry out. Dermestid larvae were characteristic of the dry stage of decay and lots were found in the muscle mass and on bones. In south-eastern Brazil, *Dermestes maculatus* is also recognized as a forensic indicator (Carvalho *et al.*, 2000).

An association has been found between evidence of the presence of dermestids, with other species, and post mortem interval. For example Arnaldos *et al.* (2005), in their succession studies in south-eastern Spain, recorded Nitidulidae and Dermestidae at the same stage of decomposition, linking their presence on the body. Post mortem interval determination is most accurate when based on evidence of the presence of several species of beetles that are normally found in association, rather than on single species of beetle alone.

**Ecology of clown beetles (Histeridae)**

This family is known to be part of the insect assemblage from the bloat stage, through the decay stages and into the dry stage. Histerid larvae and adults feed on the larvae of flies colonizing the body in these decomposition stages. Stevenson and Cocke (2000) explored the life cycle of the histerid beetle *Arcinops pumilo* (Ericson). They suggest that in laboratory-bred cultures, the adult will consume 3–24 muscid eggs per day and that the larva will consume 2–3 eggs per day in order to develop satisfactorily. According to Crowson (1981), at 20–25°C histerids beetles take 31–62 days to pass through their life cycle from egg to adult. The eggs and larvae produced at this temperature tend to be large in size.
Histerids tend to be active during the night and to hide underneath the corpse during daylight. This can account for variations in records of assemblages and the range of species present on the corpse. Equally, the decomposition stage, in which histerid beetles are present on the corpse, can vary from location to location. Korvarik (1995) found that histerid beetles arrived on the body soon after flies had colonized it. This supports the findings of Payne (1965), who recorded them during bloat, which occurred from day 1, in active and advanced decay stages, as well as in the initial dry stages of decomposition, which was recorded from day 5 onwards.

Richards and Goff (1997), investigating insect succession on pigs placed in woods at different altitudes in Hawaii, recorded *Hister noma* Erichson and *Saprinuslugens* Erichson in their collections. They too stated that histerid beetles invaded a body at the end of the bloat stage.

**Ecology of checkered or bone beetles (Cleridae)**

Members of the family Cleridae feed on carrion and are often called bone beetles. They have been classified by some workers as members of the Cornetidae rather than the Cleridae, although other researchers have retained the family name Cleridae to include such genus as *Necrobia*. Kulshrestha and Satpathy (2001) comment on the variation in the family names of these beetles. The use of the word ‘Cleridae’ for the family name has been chosen in this account, as it is a familiar term in forensic entomology.

Cleridae have been found from bloat through to the dry stage of decomposition, although the association with a particular decomposition stage may differ from country to country. For example, in the UK *Necrobia* species can be associated with dry carcasses and bone remains (Cooter, 2006). In India, Kulshrestha and Satpathy (2001) identify
Cleridae and Dermestidae as the most common beetles infesting the dry stage of decomposition of human remains. They noted the clerid *Necrobia rufipes* on remains from an environment where the average temperature was 16.5°C and the relative humidity was 71 %, although this species has also been recorded at a higher temperature and a relative humidity of 46 %. This species is called the red-legged bacon beetle, having been a noted stored product pest. It is 4–5mm long and dark blue in colour. Its legs, and the segments at the base of the antennae, are red.

Clerids may influence interpretation of the cause of death of the corpse. Members of this family, along with silphids and histerids, have been found to cause damage to the cadaver skin and these marks, at first sight, resemble gunshot wounds. Such holes serve as holes for breeding in, or result from feeding (Benecke, 2004). Therefore, care should be taken in interpreting damage on well decomposed bodies where there is evidence of the presence of members of any of these three families.

**Ecology of rove beetles (Staphylinidae)**

Rove beetles arrive on the body in the bloat stage of decomposition, or even sooner. They are predators of fly colonizers feeding on the body and they feed on both the eggs and larvae. Chapman and Sankey (1955) recorded the following species of rove beetle on rabbit carcasses: *Anotylus (= Oxytelus) sculpteratus* Gravenhorst; *Philonthus laminatus* (Creutzer); *Philonthus fuscipennis* (Mannerheim) *Creophilus maxillosus*; *Tachinus rufipes* (Degeer); *Aleochara curtula* (Goeze). These carcasses were placed within 30–40 metres of each other in shrubbery, under a plane tree or in thick meadow grass. Goff and Flynn (1991) recovered specimens of the same genus, *Philonthus* (adult *Philonthus longicornis* Stephens), from samples of sandy soil and leaf litter from beneath where a body had lain at Mokuleia, Oahu, Hawaii.
The presence of Staphylinidae will vary with the season. In spring, Centeno et al. (2002) recorded Staphylinidae on an unsheltered corpse throughout the stages of decomposition. In summer, however, staphylinids were absent from the unsheltered copse and were only recorded in a sheltered corpse during the bloat stage. In contrast, in autumn, on the unsheltered corpse, Staphylinidae were recorded in both the advanced decay and dry stages of decomposition. Their presence cannot be interpreted without considering environmental conditions such as temperature and exposure to sunlight.

**Ecology of dung beetles (Scarabaeidae)**

The Scarabaeidae are commonly known as dung beetles. Many of the dung beetle species will inhabit tunnels which they construct beneath a corpse. Two of the most common genera of Scarabaeidae are *Onthophagus* and *Aphodius* (Payne et al., 1968). As with many other beetle species, because they are not immediately obvious on a corpse, their presence can be missed.

Scarabaeidae, in a study in an urban area conducted in south-eastern Brazil, were the second most frequent colonizer on a pig carcass; the calliphorid *Chrysomaalbiceps* was the major colonizer (Carvalho et al., 2000). Three species were considered by Carvalho et al. to be important forensic indicators for post mortem determination, because they had been recovered from human cadavers, or from both human cadavers and pig carcasses, in forest environments near Campinas City, Brazil. The species were *Deltochilum brasiliensis* Castelnau, *Eurysternusparallelus* Castelnau, which were found on human cadavers, and *Coprophanaeus (Megaphanaeus)* ensifer (Germar), which was found with *Canthon* sp. and *Scybalocanthon* sp. on both pig and human corpses. Despite this association, the presence of suitable food, rather than a particular stage of
decomposition, appears to be the deciding factor in whether or not Scarabaeidae are present on a body in any geographic region.

**Objectives:**

The objectives of this study are to:

1- Record the colonization of beetles and first appearance on exposed and wrapped carcasses.

2- Determine the effect of season and temperature on forensic beetles (abundance and succession).

3- Observe the decomposition stages of carcasses.

4- Determine the effect of wrapping on forensic beetles (abundance and succession).
MATERIALS AND METHODS

This study was conducted at Khartoum State in the animal house of the Department of Zoology, Faculty of science, University of Khartoum, during winter season (December 2013 – January 2014) and summer season (May 2014 – June 2014).

Study sites

Khartoum State is situated at the center of the Sudan between latitudes (16’ 30”and 15’ 10”N) and longitudes 31’ 35” and 34’ 20”E). The State forms a rectangle which is traversed, from South to North by River Nile and its two tributaries, the White and the Blue Nile. The State is bounded from the North by the River Nile and the North State, and from the South by Gezira and the White Nile State. The East side of the State extends to Kassala and Gaderif States, while the Western wing of the State expands to North Krodofan State.

The whole State is located within the zone of the semi-arid climate, which is characterized by sporadic summer rains averaging around 150-250mm from July to September. Temperature varies from 25°C, in the winter season, to 45°C in May summer season. The soil is the clay type, with low permeability and high pH values, 8.0 upwards.
**Animal Models:**

Six sets of different animal carcasses were used in each trial as the following:

1) 2 exposed rabbits.
2) 2 wrapped rabbits.
3) 2 exposed fishes.
4) 2 wrapped fishes.
5) 2 exposed pigeons.
6) 2 wrapped pigeons.

**(1) Rabbits**

Freshly killed rabbits (*Oryctolagus cuniculus*) were used, as they are an internationally accepted substitute for human bodies (Catts & Goff 1992), each rabbit weighing approximately 1.5kg (Plate 1). Four rabbits were killed by subjecting them to smell a piece of cotton wetted in chloroform. Two of the carcasses were put exposed to insects and the other two carcasses were wrapped with three layered sheets (about 50cm) of white cloth (dabalan) and tightly tied by pieces of cloth at both ends. The rabbit was put uncovered by earth i.e. to be exposed to insects. The carcasses were then laid in wire mesh cage (2×1×2m), with larger 5cm fence wire mesh. The cages were used to protect the carcasses from large scavengers; they were in direct contact with the ground (which covered with sand soil) to allow natural ground arthropod succession. They were put in shaded area, at least 50cm apart and 100cm from the other animal carcasses (fishes and pigeons).
(2) Pigeons

Four domestic pigeons (*Columba livia domestica*) (Plate 2), were killed by subjecting them to smell a piece of cotton wetted in chloroform. Two of the carcasses were put exposed to insects and the other two carcasses were wrapped with three layers sheets about 50cm of white cloth (dabalan) and tightly tied by piece of cloth at both ends. The rabbit was put uncovered by earth i.e. to be exposed to insects, and treated similarly to the above.

(3) Fishes

Four freshly killed fishes (*Oreochromis niloticus*) (Plate 3), were also used as experimental carcasses. Two of the carcasses were put exposed to insects and the other two carcasses were wrapped with three layers sheets about 50cm of white cloth (dabalan) and tightly tied by piece of cloth at both ends, and treated similarly to the above.

Any remains of the carcasses were removed from the area after each trial.

**Sampling procedure**

The carcasses were sampled three times a day (morning, starting at 8:00, midday, starting at 13:00 and evening starting at 17:00). The time of manipulation and sampling did not exceed 10min to keep the disturbance of carcasses at minimum and maintain the integrity of micro-community. The recording and collection of the arthropods were based on the recommendations of Lord and Burger (1983); collection was done from all studies of carcasses using forceps, cups and hands with gloves. During each observation time the carcasses were described in detailed in terms of
physical appearance to determine the decomposition stage. All Coleopterans
were visually classified and put in labeled containers to preserve and identify them later; also during each visit to the carcasses the Coleopteran presence and numbers were recorded as per guide line outlined by Catt and Haskell (1990). The ambient air temperature and the photographic record of all carcasses were also maintained. During the sampling, care was taken to limit disturbance to the insects and the decomposition process, only small sample of beetles – depending on the whole number colonized on the carcass.

The Coleopteran adults were killed by freezing in zero temperature and preserved in 70% alcohol for identification later in the laboratory under the dissecting microscope by using suitable Coleopteran keys or guide books (e.g. Smith 1986, Almeida and Mise 2009, and Aballay et al. 2013).

Larvae were killed by dropping them in hot water (about 70-80°C), then preserved in 70% alcohol and stored for identification later.

**The Effect of Season on the Forensic Beetles**

These experiments were carried out during winter season (December 22\textsuperscript{th} to January 23\textsuperscript{th}) and repeated in summer season (May 10\textsuperscript{th} to June 10\textsuperscript{th}).
Plate (1) Exposed and wrapped rabbits

Plate (2) Exposed and wrapped pigeons
Plate (3) Exposed and wrapped fishes
RESULTS AND OBSERVATIONS

1. Decomposition of carcasses

Most forensic entomological studies describe the physical characteristics of the decomposition process in context with the entomological activity on the carcasses (e.g. Payne 1965, Anderson & VanLaerhoven 1996). Descriptions can include classifications into which the various stages of decomposition can be placed (Payne 1965). These descriptions were used as a guide and five decomposition stages (fresh, bloated, active decay, post-decay and skeletonization or dry) were used to best describe the process in this trial. These characteristics were found to be similar to the classes described by Anderson & VanLaerhoven (1996). However, decomposition is an ongoing process and the differences between the physical characteristics of the stages may not always be clear. For this, there are transitional stages in which characteristics from the adjoining decomposition stages can be present.

Carcasses in summer decayed much faster than those in winter. In summer the carcass reached the dry stage only in 10 days, when the average daily temperature was (25-42) °C. In contrast, during winter 14-16 days were required for the carcass to reach the dry stage, when the average daily temperature was (21-32) °C.

Fresh stage lasted less than one day in summer and whole day in winter after the exposure of the carcasses. There were no morphological changes visible on the carcass.
Bloated stage begins when bloating of carcass is first observed. This is caused by a build up of gases inside the carcass which results from anaerobic protein decomposition. This stage lasted from only one day in summer to two or three days in winter.

Active decay stage is recognizable by the skin of the corpse breaking up and starting to slough from the body. The duration of this stage varied from three or four days in summer to five or six days in winter. Towards the end of this stage, the blowfly maggots leave the carcass as pre-pupae.

Post-decay stage begins when most of the fly larvae have left the carcass and all of the internal organs are reduced. During this stage the numbers of beetles have increased. This stage lasted from four or five days in summer to six or seven days in winter.

Dry or skeletonization stage begins when no maggots remain on the carcass and lasts until carrion fauna are no longer found associated with the remains. The carcasses were in the process of drying up and most of the flesh has been removed. By the end of the experiment, only dried skin, fur, cartilage, feather, scales and bones were left of the carcasses.

The data below indicate a direct correlation between the rate of carrion decomposition and temperature in summer and winter.
Graph (1) Temperature and decomposition stages during winter trial
(December 2013 to January 2014)
Graph (2) Temperature and decomposition stages during summer trial (May 2014 to June 2014)
2. Coleoptera associated with carcasses

The first species of Coleoptera arriving during the bloat stage of decomposition included members of the families: Staphylinidae (rove beetles), Histeridae (clown beetles) adults were observed feeding on fly eggs and larvae, Cleridae (red-legged ham beetles) adults and larvae, and Dermestidae (skin beetles) adults, larvae and pupae.

Observations

1- Winter trial:

(a) Exposed rabbit

Three families, five genera and species of the order Coleoptera (as forensic insects) were collected, Cleridae: Necrobia rufipes (Plate 4), Dermestidae: Dermestes maculates (Plate 6), Histeridae: Hister sp., Euspilotus sp., Xerosprinus sp. (Plate 9).

(b) Wrapped rabbit

Three families, five genera and species of the order Coleoptera (as forensic insects) were collected, Cleridae: Necrobia rufipes, Dermestidae: Dermestes maculates, Histeridae: Hister sp., Euspilotus sp., Xerosprinus sp.

(c) Exposed pigeon

Three families, five genera and species of the order Coleoptera (as forensic insects) were collected, Cleridae: Necrobia rufipes, Dermestidae: Dermestes maculates, Histeridae: Hister sp., Euspilotus sp. and Xerosprinus sp.
(d) Wrapped pigeon

Two families, four genera and species of the order Coleoptera (as forensic insects) were collected, **Dermestidae: Dermestes maculates** and **Histeridae: Hister sp., Euspilotus sp., Xerosprinus sp.**

(e) Exposed fish

Four families, six genera and species of the order Coleoptera (as forensic insects) were collected, **Cleridae: Necrobia rufipes,** **Dermestidae: Dermestes maculates,** **Histeridae: Hister sp., Euspilotus sp., Xerosprinus sp.** and **Staphylinidae: Philonthus sp.** (Plate 11).

(f) Wrapped fish

Four families, six genera and species of the order Coleoptera (as forensic insects) were collected, **Cleridae: Necrobia rufipes,** **Dermestidae: Dermestes maculates,** **Histeridae: Hister sp., Euspilotus sp., Xerosprinus sp.** and **Staphylinidae: Philonthus sp.**

2- Summer trial:

a. Exposed rabbit

Three families, five genera and species of the order Coleoptera (as forensic insects) were collected, **Cleridae: Necrobia rufipes**, **Dermestidae: Dermestes maculates**, **Histeridae: Hister sp., Euspilotus sp., Xerosprinus sp.**
b. Wrapped rabbit

Three families, six genera and species of the order Coleoptera (as forensic insects) were collected, **Cleridae**: *Necrobia rufipes*, **Dermestidae**: *Dermestes maculates*, *Reesa vespulae* (Plate 12), **Histeridae**: *Hister sp.*, *Euspilotus sp.*, *Xerosprinus sp.*

c. Exposed pigeon

Three families, five genera and species of the order Coleoptera (as forensic insects) were collected, **Cleridae**: *Necrobia rufipes*, **Dermestidae**: *Dermestes maculates*, **Histeridae**: *Hister sp.*, *Euspilotus sp.* and *Xerosprinus sp.*

d. Wrapped pigeon

Two families, four genera and species of the order Coleoptera (as forensic insects) were collected, **Dermestidae**: *Dermestes maculates* and **Histeridae**: *Hister sp.*, *Euspilotus sp.*, *Xerosprinus sp.*

e. Exposed fish

Three families, five genera and species of the order Coleoptera (as forensic insects) were collected, **Cleridae**: *Necrobia rufipes*, **Dermestidae**: *Dermestes maculates*, **Histeridae**: *Hister sp.*, *Euspilotus sp.*, *Xerosprinus sp.*

f. Wrapped fish

Three families, five genera and species of the order Coleoptera (as forensic insects) were collected, **Cleridae**: *Necrobia rufipes*, **Dermestidae**: *Dermestes maculates*, **Histeridae**: *Hister sp.*, *Euspilotus sp.*, *Xerosprinus sp.*
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**Table (1)**Beetles on exposed rabbit in winter trial

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**Table (2)**Beetles on wrapped rabbit in winter trial
### Table (3) Beetles on exposed pigeon in winter trial

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### Table (4) Beetles on wrapped pigeon in winter trial

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<td>Dermestidae (skin beetles)</td>
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<td>Dermestes maculates</td>
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<tr>
<td>Histeridae (clown beetles)</td>
<td></td>
<td>Hister sp.</td>
<td>Euspilotus sp.</td>
<td>Xerosprinus sp.</td>
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<tr>
<td>Staphylinidae (rove beetles)</td>
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</table>
**Table (5)** Beetles on exposed fish in winter trial

<table>
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<th>Genera and species</th>
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<td>[17]</td>
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</tr>
<tr>
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<tr>
<td>Philonthus sp.</td>
<td>Staphylinidae (rove beetles)[28]</td>
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**Table (6)** Beetles on wrapped fish in winter trial

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<td>[5]</td>
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<tr>
<td>Dermestes maculates</td>
<td>Dermestidae (skin beetles)</td>
<td>[16]</td>
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</tr>
<tr>
<td>Hister sp., Euspilotus sp., Xerosprinus sp.</td>
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<tr>
<td>Philonthus sp.</td>
<td>Staphylinidae (rove beetles)[19]</td>
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<td>Family</td>
<td>Genera and species</td>
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<td>Histeridae (clown beetles)[134]</td>
<td><em>Hister sp.</em></td>
<td><em>Euspilotus sp.</em></td>
<td><em>Xerosprinus sp.</em></td>
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</table>

**Table (7)** Beetles exposed rabbit in summer trial

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<thead>
<tr>
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<th></th>
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<tr>
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<td><em>Reesa vespulae</em></td>
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<td><em>Euspilotus sp.</em></td>
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**Table (8)** Beetles on wrapped rabbit in summer trial
### Table (9) Beetles on exposed pigeon in summer trial

<table>
<thead>
<tr>
<th>Family</th>
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</tr>
</thead>
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<td><em>Euspiolus sp.</em></td>
<td><em>Xerosprinus sp.</em></td>
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<tr>
<td>Staphylinidae(rove beetles)[0]</td>
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### Table (10) Beetles on wrapped pigeon in summer trial

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<th>-</th>
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</thead>
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<table>
<thead>
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<tr>
<td>Dermestidae (skin beetles) [42]</td>
<td><em>Dermestes maculates</em></td>
<td>-</td>
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<tr>
<td>Histeridae (clown beetles) [36]</td>
<td><em>Hister sp.</em></td>
<td><em>Euspinotus sp.</em></td>
<td><em>Xerosprinus sp.</em></td>
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<tr>
<td>Staphylinidae (rove beetles) [0]</td>
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**Table (11)** Beetles on exposed fish in summer trial

**Table (12)** Beetles on wrapped fish in summer trial

(1) **Rabbits**

On the rabbits carcasses the members of the family (Histeridae) were the first observed coleopterans, which appeared and dominated both exposed and wrapped carcasses with higher frequencies in summer trial. They also recorded longer presence on the wrapped rabbit in winter trial.
The family (Dermestidae) also recorded early appearance on rabbit carcasses in both winter and summer trials. Their highest numbers were found on the exposed rabbit in winter trial, and the lowest numbers were found on the exposed one in summer trial.

The members of the family (Cleridae) generally appeared in low numbers in all trials, with little higher frequencies on both exposed and wrapped rabbits in summer trial. The family (Staphylinidae) was completely absent on all rabbit carcasses in both trials.

**Graph (3)** Frequencies of beetles on exposed rabbit in winter trial
(December 2013 to January 2014)
Graph (4) Frequencies of beetles on exposed rabbit in summer trial
(May 2014 to June 2014)
Graph (5) Frequencies of beetles on wrapped rabbit in winter trial
(December 2013 to January 2014)
Graph (6) Frequencies of beetles on wrapped rabbit in summer trial

(May 2014 to June 2014)
(2) Pigeons

The Histerids were also the first coleopterans which appeared and dominated on pigeon carcasses especially on exposed carcasses in both winter and summer trials: higher frequencies in exposed carcasses than the wrapped ones.

The members of the family (Dermestidae) were also found on all pigeon carcasses with higher frequencies on the exposed pigeon in winter trial and lower frequencies on both exposed and wrapped carcasses in summer trial especially the wrapped one.

The family (Cleridae) recorded very low appearance on all pigeon carcasses with complete absence on wrapped pigeon in winter trial. The members of the family (Staphylinidae) were completely absent on all pigeon carcasses in both trials.
Graph (7) Frequencies of beetles on exposed pigeon in winter trial

(December 2013 to January 2014)
Graph (8) Frequencies of beetles on exposed pigeon in summer trial

(May 2014 to June 2014)
Graph (9) Frequencies of beetles on wrapped pigeon in winter trial

(December 2013 to January 2014)
Graph (10) Frequencies of beetles on wrapped pigeon in summer trial

(May 2014 to June 2014)
(3) Fishes

The members of both (Hiteridae) and (Dermestidae) families dominated and recorded high frequencies on both exposed and wrapped fish carcasses in summer trial, and low frequencies of both families on wrapped and exposed fishes in winter trials.

The family (Cleridae) also recorded high frequencies in summer trial on both exposed and wrapped fishes.

The only appearance of the members of the family (Staphylinidae) was on the exposed and wrapped fishes in winter trial with completely absence in summer trial.
Graph (11) Frequencies of beetles on exposed fish in winter trial
(December 2013 to January 2014)
Graph (12) Frequencies of beetles on exposed fish in summer trial

(May 2014 to June 2014)
Graph (13) Frequencies of beetles on wrapped fish in winter trial

(December 2013 to January 2014)
**Graph (14)** Frequencies of beetles on wrapped fish in summer trial

(May 2014 to June 2014)
Plate (4) Adult of family Cleridae (*Necrobia rufipes*)

Plate (5) Larva of family Cleridae (*Necrobia rufipes*)
Plate (6) Adult of family Dermestidae (*Dermestes maculates*)

Plate (7) Larva of family Dermestidae (*Dermestes maculates*)

Plate (8) Pupa of family Dermestidae (*Dermestes maculates*)
Plate (9) Adult of family Histeridae (*Hister sp.*, *Euspirotus sp.*, *Xerosprinus sp.*).

Plate (10) Larva of family Histeridae.
Plate (11) Adult of family Staphylinidae (*Philonthus sp.*)

Plate (12) Adult of family Dermestidae (*Reesa vespulae*)
DISCUSSION

Forensic entomology is a developing field of forensic science, so there are many avenues to investigate. These avenues include novel directions that have never been addressed, as well as more critical and rigorous research into areas which have already been explored. Most research in forensic entomology has focused on flies, and beetles (Coleoptera) have been at best under-emphasized. To contextualize the neglect, throughout the world there are at least as many species of Coleoptera that may visit a particular carcass as Diptera (Braack 1986; Louw and van der Linde 1993; Bourel et al. 1999; Lopes de Carvalho et al. 2000; Pérez et al. 2005; Shea 2005; Watson and Carlton 2005; Salazar 2006; Martinez et al. 2007). A common assumption underlying the neglect of Coleoptera is that Diptera locate corpses faster, and thus give a more accurate estimate of minimum Post Mortem Interval (PMI$_{\text{min}}$). Recent observations (Midgley and Villet 2009b) have shown that Thanatophilus micans (Silphidae) can locate corpses and start breeding within 24 h of death, and thus the potential utility of estimates based on this species is equal to that of those based on flies.

Beetles form a taxonomically and ecologically diverse part of the carrion insect community (Smith 1986; Braack 1986; Bourel et al. 1999; Shea 2005; Tabor et al. 2004; Watson and Carlton 2005; Salazar 2006), thus providing a wide spectrum of sources of potential evidence. They are also integral to postmortem biology.

The species of Coleoptera increase in number both of individuals and species during advanced stages of decomposition in open environment and are absent or less represented indoors (Goff 1991). As forensic insects beetles are encountered in great numbers during the faunal succession process; moreover their biological traits may be used to estimate the post mortem interval.
According to Smith (1986) the families of Coleoptera of forensic interest are: Carabidae, Hydrophilidae, Silphidae, Leiiodidae, Staphylinidae, Histeridae, Cleridae, Anthicidae, Dermestidae, Nitidulidae, Rhizophagidae, Ptinidae, Tenebrionidae, Scarabaeidae, Geotrupidae and Trogidae.

No specific insect has been associated with a particular species of corpse, according to Hough (1897), who worked on bodies of horses, snakes, cats, dogs, fish and humans which were left exposed on the surface of the ground. Despite this lack of specific relationship, because the pig is considered biologically similar to the human, it is the preferred model for forensic entomology research on succession in humans, where human cadavers are not used or for legal reasons cannot be used.

Cleridae (Checkered beetles) is a family with about 150 genera and 4,000 species, with 61 genera and 886 species in the Neotropical region. They attack insects specially wood-boring beetles. The adults are very active specially during the day. They are often found on flowers, foliage and tree trunks.

*Necrobia ruficollis* [Fabricius 1775] and *Necrobia rufipes* [DeGeer 1775] occur in carrion and products of animal origin. *Necrobia rufipes* is predator of dipteran and coleopteran larvae. It is also associated with stored meats, such as dry fish, skin, dead animal bones, some oily seeds and stored products, mainly those with high protein indices, being also found in animal rations (Gredilha et al. 2005; Ashman 1963).

Dermestidae (Hide beetles) is a family with about 45 genera and 850 species, with 20 genera and 248 species in the Neotropical region. Also known as skin beetles, they are primarily scavengers that feed on dried skin and other soft remains of animals such as fur, feathers, scales, wool and leather. They also feed on carpets, silk, dried meats and dead insects. Some are pests of storage products such as grains, seeds, corks or cereal products.

*Dermestes maculatus* (De Geer) is a notorious pest of dried fish and fish meal, is known to damage wooden frames as well as polystyrene and glass
fiber wadding in premises when the last instar larva is about to pupate (Turner 1986; Wildey and Wayman 1979). The insect pests of dried animal products also attack living insects *Dermestes* spp. on silkworm pupae and adults (Kumar *et al.* 1988; Veer *et al.* 1996). Use of infested woolen materials can cause allergic reactions like urticarial and papulovesicular lesions in man (Ahmed *et al.* 1981).

The nature of the food is important to the success of dermestid colonization. McManus (1974) considered that the optimum rate of energy consumption for *Dermestes maculatus* was 0.17–0.28 kilocalories per gram per day. Where *Dermestes maculatus* was raised on fish with a high lipid content as a food source, a shorter length of larval stage was recorded (Obsuji, 1975). *Dermestes* spp. have been shown to require dietary sterols, including cholesterol, campesterol or 7-dehydrocholesterol to complete their life cycle (Levinson, 1962). Once the larvae have reached the pre-pupal condition, they migrate to pupate. This can result in larvae boring into a variety of substances in order to avoid cannibalism as they pupate. In addition, Dermestid larvae can delay the time of their pupation by up to 20 days if there is no suitable place to pupate (Archer and Elgar, 1998).

Histeridae (Clown beetles) is a family with about 200 genera and 3,000 species, with 139 genera and 1,047 species in the Neotropical region. They are mainly predators of soft body insects’ larvae and eggs, particularly those of Cyclorrhaphan Diptera. Most occur in carrion, dung, decomposing plant materials, such as fungi, and tree wounds. Some live under loose bark or in galleries of wood-boring insects, where they prey on other organisms. The greatly flattened species live under bark of dead or dying trees. Cylindrical species occur in tunnels of bark beetles and other wood-boring insects. Most species are neither flattened nor cylindrical and are abundant in the early stages of decay of carcasses.
This study was carried out at the Faculty of Science, Department of Zoology; nearby the insectary in small rabbit cages located in shaded area. To observe and record colonization and succession of Coleoptera species on wrapped and exposed rabbit, fish and pigeon carcasses during winter and summer seasons.

**Decomposition of carcass**

Decomposition of the bodies of dead animals is a microsuccessive process in which it is possible to distinguish several stages with their corresponding forms of carrion and the necrophage fauna proper to them. The categorization varies depending mainly on the length of the decomposing period and the type of carrion. Bornemissza (1956) included five stages of decomposition in the carrion of guinea pigs over a span of 450 d. Payne (1965) recorded five stages in piglets; however, Reed (1958) reported four stages of decomposition. Fuller (1934) used division into only three stages. Cornabay (1974) studied the carrion of toads and lizards where no stages of decomposition can be visually observed. The stages of decomposition recognised in my study follow Payne’s classification. One must keep in mind the fact that decomposition is a continuous process and discrete stages do not actually exist in nature (Schoenly and Reid 1987). The above mentioned decay stages only have a descriptive value.

Fresh stage lasted less than one day in summer and only one day in winter after the exposure of the carcasses. There were no morphological changes visible on the carcass.

Bloated stage begins when bloating of carcass is first observed. This is caused by a build up of gases inside the carcass which results from anaerobic protein decomposition. This stage lasted from only one day in summer to two or three days in winter.
Active decay stage is recognizable by the skin of the corpse breaking up and starting to slough from the body. The duration of this stage varied from three or four days in summer to five or six days in winter. Towards the end of this stage, the blowfly maggots leave the carcass as pre-pupae.

Post-decay stage begins when most of the fly larvae have left the carcass and all of the internal organs are reduced. During this stage the numbers of beetles have increased. This stage lasted from four or five days in summer to six or seven days in winter.

Dry or skeletonization stage begins when no maggots remain on the carcass and lasts until carrion fauna are no longer found associated with the remains. The carcasses were in the process of drying up and most of the flesh has been removed. By the end of the experiment, only dried skin, fur, cartilage, feather, scales and bones were left of the carcasses.

These data are consistent with the average daily temperatures recorded in each season. These observations can be attributed to differences in breeding biology of blowflies. The development of blowfly larvae, which are responsible for the process of skin puncturing that ended the bloated stage earlier by passing to aerobic decomposition (decay stage), is faster in the warmer season (Ash and Greenberg 1975, Catts and Goff 1992 and Greenberg 1991). Tantawi et al. (1996) noted, that the period of the bloated stage depends more on the number of larvae infesting the carcass rather than on temperature. During the decay stage, temperature plays a primary role in the length of the stage—the carcass is open and its flesh dries rapidly especially in hot weather.

The results of arthropod exclusion experiments demonstrate accelerated breakdown and decay of rabbit carrion in the presence of necrophagous and saprophagous insects. Payne (1965) and Abell et al. (1982) obtained the same results.
Differences in the microclimate, depending on the position of carcasses relative to trees and shrubs, obviously influenced the rate of decay (Bornemissza 1956). Rates of decomposition also vary with the temperature (Doenier 1940), which especially affects bacterial activity (Putman 1983) and developmental rates of carrion frequenting insects (Ash and Greenberg 1975).

**Coleoptera succession**

The activity of some beetles started after the first generation of flies have come and gone. Six species in four families of coleoptera were collected in this study, Histeridae, Cleridae, Dermestidae, and Staphylinidae, that agrees with Gurafi (2013) who observed the presence of *Hister sp.* (Histeridae), *Dermestes maculates* (Dermestidae), *Necrobia rufipes* (Cleridae). Most of the species found are members of the family Histeridae. Aly (2013) also found2 families of Coleoptera: Dermestidae and Histeridae in Egypt. Dupont (2011) mentioned that among the insects assessed were Coleoptera: Cleridae, Curculionidae, Dermestidae, Histeridae, Mordellidae, Ptiliidae, Scarabaeidae, Silphidae, Staphylinidae (subfamily Tachyporinae), Trogidae in Cameroon. The family, Histeridae, is very diverse, so different species can be found on the body at different times because of their various feeding habits.

In these experiments three different animals were used: rabbits, fishes and pigeons, and in all experiments the carcasses were either left exposed or wrapped. In winter trial members of the family (Histeridae) were the first observed coleopteran to appear and dominate both exposed and wrapped carcasses with high frequencies on the exposed rabbits and low frequencies on both exposed and wrapped pigeon. The family (Dermestidae) was also present in bloated stage (adults) until the dry stage of the decomposition (adults, larvae and pupae). They were found in large numbers on the exposed pigeon and
rabbit, beside low numbers on both wrapped pigeon and fish. The adults of the family (Cleridae) were observed in low numbers on all carcasses generally, with completely absence on the wrapped pigeon. Cleridae recorded larger numbers in both exposed and wrapped fishes. The family (Staphylinidae) was observed only on exposed and wrapped fishes.

In summer trial recorded the same beetles as winter trial with large numbers of the family Histeridae especially on both exposed and wrapped rabbits. The members of the family Dermestidae were found in higher frequencies on exposed rabbit, pigeon and fish more than the wrapped ones. The family Cleridae also recorded high frequencies on both exposed and wrapped fish carcasses and low frequencies on exposed and wrapped rabbit and pigeon carcasses.

The members of the family Staphylinidae were completely absent in summer trial.

In these experiments the insects and their succession were the same as insects colonizing the exposed carcasses, that agrees with Gurafi (2013, 2014), Who observed the presence of *Chrysomya albiceps* (Calliphoridae), *Sarcophaga tibialis* (Sarcophagidae), *Megaselia scalaris* (Phoridae), *Hister spp* (Histeridae), *Dermestes maculates* (Dermestidae), *Necrobia rufipes* (Cleridae). Kelly (2006) observed the same coleopteran families in South Africa (Dermestidae, Cleridae, Histeridae and Silphidae) in both exposed and wrapped pigs during summer and winter seasons.

The sequence of Coleoptera succession observed in this study follows the same general patterns found in both temperate and tropical areas (Bornemissza (1956), Ash and Greenberg (1975), Cornabay (1974), Erbeling and Erbeling (1986), Fuller (1934), Johnson (1975), Kentner and Streit (1990), Nabaglo
(1973), Payne (1965), Peschke et al. (1987), Reed (1958), Richards and Goff (1997), Tantawi et al. (1996), and Wasti (1972)).
Conclusion

The aims of this study were to determine the influence of a) season and b) wrapping on different carcasses decomposition and Coleoptera succession.

A) Effect of season
B) Effect of wrapping

(1) The entomofauna of cadavers and succession studies are newly developing fields in Sudan and need more attention from scientists.

(2) This is the first study done in Sudan on the Coleopteran fauna of carcasses. Data acquired from the study could be used to help interpret the Coleopteran evidence in forensic cases in Khartoum and elsewhere in the future.

(3) The succession of insects on rabbit, fish and pigeon carcasses can, in general, be considered to be featured by the members of two orders, Diptera and Coleoptera.

(4) In this study, Coleoptera were targeted as forensic insects. Families, genera and some species arriving on these carcasses were identified. Records were done in summer and winter seasons.
Recommendations

(1) Illustrate the importance of forensic beetles in legal investigations.

(2) Activate beetles laboratory in the General Administration of Criminal Labs.

(3) The work will be more elaborate and productive if it is done for more animal models.

(4) More studies in the field of forensic insects generally and beetles specially are needed.
REFERENCES


Appendix (1)
Rabbit Decomposition stages

Plate (1) Fresh stage

Plate (2) Bloated stage
Plate (3) Active decay stage

Plate (4) Post-decay stage
Plate (5) The beginning of dry stage

Plate (6) Skeletonization or dry stage

Appendix (2)
Decomposition of wrapped rabbit

Plate (7) Rabbit carcass before wrapping

Plate (8) Rabbit carcass after wrapping

Plate (9) Wrapped rabbit carcass after 25 days

Appendix (3)
Decomposition of pigeon

Plate (10) Fresh pigeon carcass

Plate (11) Exposed pigeon carcass after 25 days

Plate (12) Wrapped pigeon carcass after 25 days

Appendix (4)

Decomposition of fish
Plate (13) Fresh fish carcass

Plate (14) Exposed fish carcass after 25ays

Plate (15) Wrapped fish carcass after 25days

Appendix (4)

Beetles
Plate (16) Histeridae (predators) feeding on fly larva (a) and (b)

Plate (17) Sampling from carcasses (a) and (b)